Trophic interactions as drivers of secondary succession in tropical rainforests: an experimental test in New Guinea

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Experimental study of mechanisms maintaining tropical diversity

Tropical rainforests, comprising typically 250–1,100 woody species per 50 ha, are the most species-rich terrestrial ecosystems on Earth[1]. Search for ecological mechanisms enabling the coexistence of hundreds of plant species remains a key mission of tropical ecology[2]. Rainforest vegetation supports complex food webs, comprising pathogens, herbivores and predators. We have estimated that 200 plant species hosted 9,600 herbivorous insect species, involved in 50,000 distinct trophic interactions in a New Guinean forest[3], and that there were 25,000 species of arthropods in 6,000 ha of rainforest in Panama[4]. Tropical plant diversity is clearly supporting a high diversity of herbivores, often specialized to certain plant lineages[5].

Interestingly, the causality of the plant-insect herbivore relationships could also go in the opposite direction, with herbivores driving plant diversity in tropical forests. Specialized herbivores and other natural enemies of plants, including fungal pathogens, could facilitate the coexistence of plant species through density dependent plant mortality. This mechanism, known as the Janzen-Connell hypothesis, was proposed 46 years ago[6,7]. It has remained at the centre of ecological theory ever since. The signature of density-dependent mortality has been repeatedly identified in spatial patterns of plant dynamics and distribution in tropical forests[8]. Rather surprisingly, it has taken >40 years before this ecological hypothesis has been tested experimentally[9,10]. Suppressing fungi reduced the diversity of seedlings; suppressing insect herbivores increased seedling survival and changed their species composition. As one of the study's title said, "pathogens and insect herbivores drive rainforest plant diversity and composition"[10]. Recently, experiments have shown that insect herbivores can also act as a density-dependent source of mortality in mature tropical forests[11].

The history of testing the Janzen-Connell hypothesis illustrates the importance of community-wide experiments, particularly in tropical forests with complex food webs[12]. We have documented >6,800 distinct plant-herbivore interactions in lowland rainforest in New Guinea, from a total of ~50,000 likely present there[3]. As noted by Lewinsohn[13], this food web, although incomplete, "is likely to be as good as it gets". We suggest that further understanding of tropical biodiversity requires experimental manipulation of such complex food webs, rather than their continued inventory.

There is a long tradition of community experiments in temperate grasslands, starting from the Rothamsted experiments in 1843 and continued later elsewhere (e.g., Cedar Creek LTER site, the Jena Experiment)[40]. Mooney[15] noted: "We have done the easy stuff, working experimentally with herbaceous communities .... we now must move on to address those ecosystems that control a good portion of the carbon, nutrient and water balances of the earth - the forests". The grassland experiments were up-scaled to tropical forests in the Sabah Biodiversity Experiment where dipterocarp diversity was varied in experimental plots[14]. Interestingly, no such experiments seem to focus on ecological succession in forests, despite the importance of forest regeneration for the maintenance of tropical biodiversity and ecosystem services[16]. This omission is particularly surprising considering the feasibility of such experiments in the tropics: early succession plots can be smaller than those required for mature forests, and tropical succession progresses rapidly, making the experiment a feasible proposition for three-year cycles of research funding.

The rainforest studies have been progressing along two trajectories, (i) producing increasingly detailed resolution of complex plant-herbivore food webs, while lacking information on the impact of these herbivores on plants[3,5], or (ii) manipulating herbivores or plant pathogens and measuring their impact on plants, but treating these trophic levels as black boxes, lacking information on the species and interactions responsible for the observed effects[9,10]. The next step, proposed here, should combine the rigor of experimental manipulation with understanding of underlying mechanisms afforded by studies on species-level resolution.

Objectives of research

We test the importance of biotic interactions as drivers of secondary succession in tropical rainforests by a series of replicated experiments manipulating successional food webs in order to uncover the effect of (i) fungal pathogens and mutualists, (ii) invertebrate herbivores, and (iii) invertebrate and vertebrate predators on the trajectory of plant succession. These experiments will take place along an elevation gradient providing natural variation in the intensity of biotic interactions. We will use them to test the following hypotheses:

H1. The course of secondary succession is partly determined by natural enemies of plants, including insect herbivores and fungal pathogens. Their removal will change the species composition and increase species diversity of the vegetation as predicted in H2 and/or H3.

H2. There is a trade-off between growth rate and anti-herbivore/fungal defense in pioneer plant species. Insect herbivores and fungi cause higher mortality in fast growing and poorly defended plant species, thus impacting the species composition and ecological trait distribution in successional vegetation.

H3. Insect herbivores, particularly generalist species, and fungal pathogens, cause density-dependent mortality in early succession stages by disproportionately attacking the most abundant plant species, increasing thus the diversity of early successional vegetation.

H4. Locally diverse plant lineages, such as species-rich genera, suffer higher mortality caused by herbivores and fungal pathogens than phylogenetically isolated plant species, including alien species. This increases phylogenetic diversity in secondary vegetation and facilitates the establishment of alien species.

H5. The impact of herbivores on secondary succession is controlled by predators, particularly ants, birds and bats, and intensifies in their absence.

H6. The importance of trophic cascades controlling secondary succession decreases with altitude. This leads to increasing importance of plant recruitment limitation and inter-specific competition, making species' growth rates and seed abundances in the soil seedbank increasingly better predictors of their success in secondary succession.

H7. The effect of trophic cascades decreases in the course of ecological succession; they have highest impact on seeds and seedlings, dominating early succession stages, while competition among plants becomes more important in later succession stages.

Methods

Experimental design. We have designed two types of experiments: a garden experiment and a seedling tray experiment. The garden experiment comprises one control and four treatment plots: T1: reduction of fungi (pathogens and mutualists), T2: reduction of insects, T3: reduction of key predators, and T4: addition of generalist herbivores with reduction of key predators (Fig. 1). We will use blocks of five 5 x 5 m plots, each randomly assigned particular treatment or control. There will be six blocks, separated by at least 100 m, established at each of the three study sites, resulting in 5 x 6 x 3 = 90 experimental plots. Each block will be initiated in a recently abandoned gardening area in a landscape with a mosaic of primary and secondary forests, and will be run for two years. The vegetation will be surveyed non-destructively every four months, and it will be harvested, with its insect communities, at the end of the experiment.

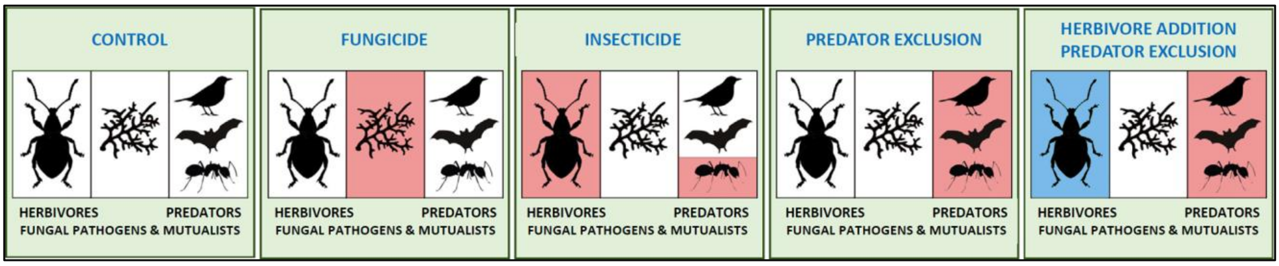

Fig. 1. Schematic representation of one block of 5 x 5 m plots in a garden experiment. The manipulation (red: reduction, blue: addition) on three target groups (insect herbivores, insect & vertebrate predators, and fungal pathogens & mutualists) are shown for the control and four treatments.

Fig. 2. Pilot experiment testing our methods on 5 x 5 m gardens in Wanang. A: start of the experiment, B: another block two months into the experiment; C: the effect of insecticide treatment (upper half of the image) can be clearly seen, D: example of pioneer vegetation after 2.5 years of succession; E: Palatability (the proportion of feeding in 50 no-choice experiments) of 62 common plant species to *Oribius* sp. 1, an extreme generalist that will be used to increase herbivore density in the lowland garden experiment.

Facilities

The project is based at **the Biology Centre of the Czech Academy of Sciences** (BC, www.bc.cas.cz/en) and **the New Guinea Binatang Research Center** (BRC, www.entu.cas.cz/png).

BRC hosts the study in PNG. It is the best-equipped and the most productive institution for biological research in New Guinea (Fig. 3). BRC includes four buildings (250 m^2 of lab space, accommodation for 35 people, a 50 kVA diesel generator, autonomous water supply), four 4WD Toyota Landcruiser vehicles, a broadband internet access via satellite link, an insect rearing facility, reference collections of plants (~1,000 spp.) and insects (~5,000 spp.) with storage space for plant and insect specimens (capacity 15,000 plant and 50,000 insect specimens) and freezers for sample storage.

Fig. 3. BRC has a team of paraecologists and students, facilities including insect imaging, herbarium and insect collections, and manages field stations in Wanang (pictured) and along the CART transect.

Significance and broader impacts

(1) This is probably the first experimental test of the importance trophic interactions have for the secondary succession in tropical forests, examining the impact of fungal pathogens and mutualists, as well as herbivores and their predators, on the vegetation dynamics. Rainforest regeneration and secondary succession is becoming increasingly important in human-impacted landscapes, as the share of early successional stages increases in tropical forests[16].

(2) The forest regeneration is studied along an altitudinal gradient, providing an opportunity **to replicate experiments along a natural gradient of decreasing herbivory and predation pressures.** This natural variability will be combined with experimentally manipulated strength of trophic interactions at each elevation.

(3) We combine manipulative experiments with detailed knowledge of plant-animal interaction webs, based on our extensive surveys of these webs in the past 20 years at our study sites. **This will allow us to combine two, so far mostly separate, research approaches to complex rainforest ecosystems: rigorous experimental approach with understanding of trophic interactions on the species level.**

References

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